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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/617,060	07/10/2003	Peter H. Kay	47675-52	2645

22504 7590 01/05/2006

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EXAMINER	
GOLDBERG, JEANINE ANNE	

ART UNIT	PAPER NUMBER
1634	

DATE MAILED: 01/05/2006

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/617,060	KAY, PETER H.	
	Examiner	Art Unit	
	Jeanine A. Goldberg	1634	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *05 December 2005*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-18* is/are pending in the application.

4a) Of the above claim(s) *18* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-17* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date *9/03; 12/03*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

1. This action is in response to the papers filed December 5, 2005. Currently, claims 1-18 are pending.

2. Claim 18 has been withdrawn as a non-elected claim.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

3. Claims 1-6, 14-17 are rejected under 35 U.S.C. 103(a) as being unpatentable over Elsas, II et al. (US Pat. 6,207,387, March 27, 2001) in view of either Ehrlich et al. (Biochimica et Biophysica Acta, Vol. 395, pages 109-119, 1975) or Hua et al. (Gov. Rep. Announce. Index US, Vol. 88, No. 18, Abstract No. 847,050 1988) and in further view of either Tyagi et al (US Pat. 6,150,097, November 2000) or Coull et al (US Pat. 6,355,421, March 2002).

Elsas, II et al. (US Pat. 6,207,387, March 27, 2001) teaches detecting mutations in genes by determining the melting temperature of the hybrid of the amplified DNA and the specific oligonucleotide (col. 10, lines 20-25). Elsas teaches that "under identical conditions, two strands that are not exactly complementary, differing by even one nucleotide, will be less stable and will dissociate at a temperature which exactly

complementary hybrids remain paired (col. 8, lines 50-57). Elsas teaches that the melting temperature between a mismatched hybrid will denature at a lower temperature than a exact matched hybrid (col. 10, lines 37-40). Elsas also teaches that fluorescence energy transfer is a specific application of this approach (col. 10, lines 49-50). Elsas teaches the different melting temperatures for the allele specific probe and detection of fluorescence (col. 11).

While Elsas teaches detecting different nucleic acids based upon melting temperature, Elsas does not specifically teach the structure of fluorescence energy transfer and does not teach using the fluorescence energy transfer for detecting methylation.

Erlich teaches Xanthomonas phage XP-12 DNA containing 5-methylcytosine completely replacing cytosine, has the highest reported melting temperature for any naturally occurring DNA (abstract). The melting temperature is 6.1 degrees Celsius higher than normal DNA containing the same percentage of adenine plus thymine (abstract), page 114). As seen in Figure2, the XP-12 DNA has a higher melting temperature (page 114).

Similarly, Hua teaches that the melting temperature of methylated Z-DNA is 387K which is 7K higher than a similar calculation for unmethylated B-DNA which is in agreement with observation (abstract).

Moreover, Tyagi et al. (herein referred to as Tyagi) teaches using nucleic acid hybridization probes having a first conformation when not interacting with a target and a second conformation when interacting with a target and having the ability to bring a

label pair into touching contact in one formation but not the other (abstract). Tyagi teaches using quenching molecules and other fluorophores as efficient quenching moieties for fluorophores when attached to nucleic acid hybridization probes (col 3, lines 40-43)(limitations of Claim 2). The probes of Tyagi contain a hairpin structure which comprise single stranded loop of the hairpin and two arm sequences which form a double stranded stem hybrid (col. 5, lines 10-15). Tyagi teaches that the molecular beacon probes may have target recognition sequences 7-140 nucleotides in length (col 5, lines 24-25)(limitations of Claims 3-6). Additionally Tyagi teaches the arms that form a stem hybrid or stem duplex are 3-25 nucleotides in length (col. 5, lines 26-27)(limitations of Claim 16). Tyagi teaches a kit which contains a hairpin probe with labels (col 19, lines 66-67, Claim 12)(limitations of Claim 18).

Coull et al. (herein referred to as Coull) teaches methods of detecting target sequences using a probe which has a measurable change in one property of at least one donor or acceptor moiety of the probe which can be used to detect, identify or quantitated the target sequence in a sample. As seen in Figure 11, configuration III, a probing segment is flanked on either side by a arm segment and either a quencher and fluorophore. The hairpin loop and stem structure allows energy transfer between donor and acceptor moieties linked at opposite ends of the nucleic acid polymer (col. 7, lines 30-37). The probing segments is designed to hybridized to at least a portion of a target sequence (col. 8, lines 35-36). In the method of Coull, a sample is contacted with the molecular beacon and a change in detectable signal associated with at least one donor or acceptor moiety of the probe is detected, identified or quantitated. Coull teaches that

the assay may be used to detect a target sequence which is specific for a genetically based disease including cancer. Coull teaches that the probing sequence hybridizes to the entire target sequence (col. 16, lines 20-25). The probing sequence will generally have a length between 5-30 units in length (col. 16, lines 32-34)(limitations of Claims 3-6). The arm segments are 2-6 units in length (limitations of Claim 16). Coull teaches that shorter probes are less costly to synthesize, are generally easier to purify and should exhibit few non-specific interactions since they will comprise less nucleobase sequence diversity (col. 20, lines 29-32)(limitations of Claim 15). Coull teaches kits which comprise one or more PNA Molecular Beacons (col. 24, lines 51-67)(limitations of Claim 18). Coull teaches considerable analysis of the Tm melting temperature for the stem-loop hairpin probes (col. 20, 37-44). Coull teaches that the probes exhibit a low inherent noise (background) and an increase in detectable signal upon binding of the probe to a target sequence (col. 7, lines 40-42).

Therefore, it would have been prima facie obvious to one of ordinary skill in the art to have modified the method of Elsas for detecting different nucleic acids based upon different hybridization properties including melting temperatures with the teachings of either Tyagi or Coull which discuss and describe stem-loop and fluorescence energy transfer with the teachings of either Ehrlich or Hua teaching the properties of methylated DNA. Detecting nucleic acids based upon different melting temperatures and dissociation properties was used to identify mismatches in nucleic acids. Both Ehrlich and Hua teach that methylated DNA and unmethylated DNA have different melting temperatures. The ordinary artisan would have recognized based upon the teachings

that in the art, namely Ehrlich and Hua, that in addition to mismatched DNA, methylation could also be detected based upon different melting temperature and dissociation rates. Combining the teachings of Elsas and either Ehrlich or Hua would yield an assay which would detect methylated nucleic acids as compared to unmethylated nucleic acids. Detection of methylated nucleic acids as compared to unmethylated nucleic acids is of interest to the clinical diagnostics because numerous genes are methylated in cancer as compared to unmethylated in normal tissue. Therefore, combining the teachings of Elsas and Ehrlich or Hua, a hybridization assay for differentiation methylated DNA from unmethylated DNA does not require the use of enzymes, solid supports would facilitate the detection of methylation in genes and may be used as an indicator for cancer. Moreover, the use of hairpin stem-loop probes with fluorophores and quenchers for detecting target nucleic acids in samples is taught by both Tyagi and Coull would have provided a fluorescent detection assay which is easily detectable in a single tube which does not require subjection to a gel or solid support. The use of FRET allows for the direct detection of nucleic acid target sequences without the requirement that labeled nucleic acid hybridization probes or primers be separated from the hybridization complex prior to detection (Coull et al. col. 1, lines 45-50). Therefore, using the specific teachings about fluorescence energy transfer techniques, as described in Coull and Tyagi, would facilitate the fluorescence energy transfer method for detecting different nucleic acids as taught by Elsas. Therefore, given all of the teachings well known in the art, at the time the invention was made, a FRET-like method for detection of different nucleic acids based upon the know property that methylated and

unmethylated DNA molecules have different hybridization properties would have been obvious to the ordinary artisan.

Response to Arguments

The response traverses the rejection. The response asserts that the examiner has failed to make out a prima facie case of obviousness. The response asserts that the mere observation by Ehrlich and Hua that melting temperatures of methylated DNA sequences can be higher than melting temperatures of non-methylated DNA sequences is not a teaching or suggestion to modify the method of Elsas from detecting nucleic acid substitutions in a sample to detecting methylated nucleic acids in a sample. This argument has been reviewed but is not convincing because the art at the time recognized that differences in nucleic acids may be characterized by different melting temperatures as exemplified by Elsas. Thus, the ordinary artisan would have been motivated to have detected additional differences in nucleic acids using the same technique of melting temperature properties, such as differences in methylated and nonmethylated nucleic acids.

The response asserts that the skilled artisan would not have any reasonable expectation that the two different types of mutations would have the same melting behavior. This argument has been thoroughly reviewed, but is not found persuasive because both Erlich and Hua teach the differences in melting temperature of methylated and unmethylated DNA. Thus, the ordinary artisan would have had a reasonable expectation of success to use the different melting behavior of methylated and unmethylated DNA to distinguish between the nucleic acids.

The response asserts that non of the references teach the element of detecting methylated nucleic acids or the element of an oligonucleotide sequence containing a region that is susceptible to methylation. This argument has been thoroughly reviewed, but is not found persuasive because the combination of the references which demonstrate detection of nucleic acids based upon different melting temperatures, the observation that methylated and unmethyalted nucleic acids have different temperatures suggests that methylated nucleic acids may be detected based upon different melting temperatures.

The response asserts that "beyond a wish for cancer diagnosis, the examiner points neither to any specific passages in the cited references which provide motivation to combine those references". This argument has been thoroughly reviewed, but is not found persuasive because assays which are able to determine a predisposition to cancer, onset of cancer and such are of real concern to the scientific community. The examiner is unclear why the motivation to detect nucleic acids by detecting differences in methylation to enable assessment of cancer is in sufficient motivation for a method.

Thus for the reasons above and those already of record, the rejection is maintained.

4. Claims 7, 10, 12, 13 are rejected under 35 U.S.C. 103(a) as being unpatentable over Elsas, II et al. (US Pat. 6,207,387, March 27, 2001) in view of either Ehrlich et al. (Biochimica et Biophysica Acta, Vol. 395, pages 109-119, 1975) or Hua et al. (Gov. Rep. Announce. Index US, Vol. 88, No. 18, Abstract No. 847,050 1988) and in further

view of either Tyagi et al (US Pat. 6,150,097, November 2000) or Coull et al (US Pat. 6,355,421, March 2002) as applied to Claims 1-6, 14-19 above, and further in view of Herman et al. (US Pat. 6,265,171, July 2001).

The combination of Elsas, Ehrlich or Hua and Tyagi or Coull does not specifically teach detecting methylation in GSTpi or calcitonin which is differentially expressed in cancer versus a normal state.

However, Herman et al. (herein referred to as Herman) teaches numerous genes which are differentially methylated at CpG islands in neoplastic versus normal tissue (limitations of Claim 7). These genes include GSTpi and calcitonin (limitations of Claims 10, 12). Herman also teaches that CpG island differential methylation may be detected in prostate cancer (col. 112, Claim 12)(limitations of Claim 12). Aberrant methylation in the 5' promoter of E-cadherin is prostate, breast and many other carcinomas (col. 27, lines 5-10).

Therefore, using the method of Elsas, Ehrlich or Hua and Tyagi or Coull in view of the teachings of differential methylation in gluthathione-S-transferase-II(pi) and calcitonin. The ordinary artisan would have been motivated to have detected methylation in these two specific genes because Herman teaches that they contain methylated CpG neoplastic versus normal tissue.

Response to Arguments

The response traverses the rejection. The response asserts that since the combination of the references fails to render independent claim 1 obvious the combination does not render the claims obvious. The response asserts that Herman

only teaches "the desire to detect methylation of CpG because the presence of methylated CpG in a 5' regulatory region can be indicative of a cell proliferative disorder." This argument has been reviewed but is not convincing because the Herman has only been relied upon to the extent that the response agrees with the examiner. As provided above, the rejection of Elsas, Ehrlich or Hua and Tyagi or Coull has been maintained, therefore, the ordinary artisan would have been motivated to have used the method to have detected differential cancer methylation as taught by Herman. Thus for the reasons above and those already of record, the rejection is maintained.

5. Claims 7, 8 are rejected under 35 U.S.C. 103(a) as being unpatentable over Elsas, II et al. (US Pat. 6,207,387, March 27, 2001) in view of either Ehrlich et al. (Biochimica et Biophysica Acta, Vol. 395, pages 109-119, 1975) or Hua et al. (Gov. Rep. Announce. Index US, Vol. 88, No. 18, Abstract No. 847,050 1988) and in further view of either Tyagi et al (US Pat. 6,150,097, November 2000) or Coull et al (US Pat. 6,355,421, March 2002) as applied to Claims 1-6, 14-19 above, and further in view of Kay et al (Leukemia and Lymphoma, Vol. 24, pages 211-220, 1997).

The combination of Elsas, Ehrlich or Hua and Tyagi or Coull does not specifically teach detecting methylation in Myf-3 which is differentially expressed in cancer versus a normal state.

However, Kay et al. (herein referred to as Kay) teaches the Myf-3 gene is abnormally hypermethylated in non-Hodgkins lymphoma (abstract).

Therefore, using the method of Elsas, Ehrlich or Hua and Tyagi or Coull in view of the teachings of differential methylation in gluthathione-S-transferase-II(pi) and calcitonin. The ordinary artisan would have been motivated to have detected methylation in these two specific genes because Herman teaches that they contain methylated CpG neoplastic versus normal tissue.

Response to Arguments

The response traverses the rejection. The response asserts that since the combination of the references fails to render independent claim 1 obvious the combination does not render the claims obvious. The response asserts that Kay only teaches "that the Myf-3 gene can by hypermethylated and that the hypermethylated status of the Myf-3 gene may provide new diagnostic indicators of malignancy." This argument has been reviewed but is not convincing because the Herman has only been relied upon to the extent that the response agrees with the examiner. As provided above, the rejection of Elsas, Ehrlich or Hua and Tyagi or Coull has been maintained, therefore, the ordinary artisan would have been motivated to have used the method to have detected differential cancer methylation as taught by Herman. Thus for the reasons above and those already of record, the rejection is maintained.

Conclusion

6. No claims allowable over the art.

7. The prior art made of record and not relied upon is considered pertinent to applicant's disclosure.

A) Collins (J. Mol. Biol. Vol. 198, pages 737-744, 1987) teaches that base modifications such as methylation are known to affect the melting temperature of DNA and may be evaluated using electrophoresis.

B) Leng et al. (Biochimica et Biophysica Acta Vol. 174, pages 574-585, 1969) teaches in comparison to native DNA, the melting temperature of methylated DNA is decreased.

C) Yamasaki et al. (Proc. Japan Acad. Vol. 74, Ser. B, pages 210, 1998) teaches methylation of four adenine bases in a decamer DNA duplex decreased the melting temperatures by 9.4 degrees.

8. Any inquiry concerning this communication or earlier communications from the examiner should be directed to examiner Jeanine Goldberg whose telephone number is (571) 272-0743. The examiner can normally be reached Monday-Friday from 7:00 a.m. to 4:00 p.m.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Jones, can be reached on (571) 272- 0745.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

The Central Fax Number for official correspondence is (571) 273-8300.

J. Goldberg

Jeanine Goldberg
Primary Examiner
December 20, 2005